High frequency of partial *SPG4* gene deletions in hereditary spastic paraplegia

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PCR-based screening detects *SPG4* mutations, usually affecting one or a few bases, in ~40% of autosomal dominant hereditary spastic paraplegia (AD-HSP) cases. Several screens, however, have failed to reveal a causative alteration despite strong linkage to *SPG4*. Our previous identification of a multi-exonic deletion in one such pedigree led us to search for HSP-associated copy number alterations more systematically. To this end we developed a multiplex ligation-dependent probe amplification (MLPA) kit which targets all *SPG4* exons as well as several exons of *SPG3A*, another gene frequently mutated in AD-HSP.

In an initial screen on 18 HSP patients, some of which had previously been screened negative for *SPG4* mutations, we identified 3 aberrant MLPA profiles. In one of the respective DNAs we found a known 3bp deletion which lies within the probe binding sequence whereas the other two harbour novel deletions of 3 adjacent *SPG4* exons each; the latter findings were subsequently validated by breakpoint mapping. In an ongoing multi-center study we are currently investigating DNA from another 53 AD-HSP index patients, all of which had been regarded *SPG4* mutation-negative based on conventional screening. Aberrant MLPA profiles have been found in at least 11 cases (21%); ten of these are consistent with additional, mainly private, large SPG4 deletions whereas one suggests an amplification of a single *SPG3A* exon. Validation of these results by independent methods as well as analysis of segregation with the disease are underway.

These preliminary data suggest partial *SPG4* deletions to be present in 10-15% of AD-HSP cases thereby totalling the involvement of *SPG4* in this disease to >50%. We believe that the kit introduced here will become a valuable tool in the molecular diagnosis of HSP.